

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application.

1-5. (Cancelled)

6. (Currently amended) A method of ~~introducing a nucleic acid molecule into the genome of~~ producing an avian species comprising a heterologous gene, comprising the steps of:

(a) providing a population of isolated sex-determined gonadal cells from avian embryos;

(b) transfecting said population with a heterologous nucleic acid molecule;

(c) transferring said population of step (b) to a fertilized recipient avian egg; and

(d) incubating said recipient egg under conditions that allow the development and hatching of said egg, thereby producing an avian species that expresses said heterologous gene ~~contacting a population of isolated gonadal cells obtained from a chick embryo with said nucleic acid molecule to yield transfected gonadal cells, and transferring said transfected gonadal cells to a fertilized recipient avian egg.~~

7. (Original) The method of claim 6, wherein said population comprises at least 0.5% primordial germ cells.

8. (Original) The method of claim 6, wherein said population comprises at least 1% primordial germ cells.

9. (Original) The method of claim 6, wherein said population comprises at least 50% primordial germ cells.

10. (Original) The method of claim 6, wherein said population comprises at least 90% primordial germ cells.

11. (Original) The method of claim 6, wherein said chick embryo is at an embryonic stage of greater than 27.

12. (Original) The method of claim 6, wherein said chick embryo is at an embryonic stage of 29-36 of gestation.

13. (Currently amended) The method of claim 6, wherein said transfected gonadal cells and said fertilized avian egg are ~~derived~~ from the same species.

14. (Currently amended) The method of claim 6, wherein said transfected gonadal cells and said fertilized avian egg are ~~derived~~ from different species.

15. (Currently amended) The method of claim 6, wherein said fertilized recipient avian egg is between stage 7-8.

16. (Currently amended) The method of claim 6, wherein said fertilized recipient avian egg is between stage 13-19.

17. (Original) The method of claim 6, wherein the breed of said chick embryo is White Leghorn.

18. (Original) The method of claim 6, wherein the breed of said chick embryo and the breed of said fertilized recipient egg are different.

19. (Currently amended) The method of claim 6, wherein said fertilized recipient avian egg is partially sterilized prior to transferring said ~~transfected gonadal~~ selected cells.

20. (Currently amended) The method of claim 6, wherein said fertilized recipient avian egg is contacted with busulfan prior to transferring said ~~transfected gonadal~~ selected cells.

21. (Original) The method of claim 6, wherein said transfected gonadal cells are transferred directly into the germinal crest of said fertilized recipient avian egg.

22. (Original) The method of claim 6, wherein the sex of said gonadal cells and the sex of an embryo in said fertilized recipient avian egg is the same.

23-25. (Cancelled)

26. (New) The method of claim 6, wherein said population of sex-determined gonadal cells comprises male gonadal primordial germ cells and less than 20% female gonadal primordial germ cells.

27. (New) The method of claim 26, wherein said population of sex-determined gonadal cells comprises male gonadal primordial germ cells and less than 10% female gonadal primordial germ cells.

28. (New) The method of claim 6, wherein said population of sex-determined gonadal cells comprises female gonadal primordial germ cells and less than 20% male gonadal primordial germ cells.

29. (New) The method of claim 28, wherein said population of sex-determined gonadal cells comprises female gonadal primordial germ cells and less than 10% male gonadal primordial germ cells.

30. (New) The method of claim 6, wherein step (d) further comprises contacting said recipient egg with a hormone.

31. (New) The method of claim 30, wherein said hormone is testosterone.

32. (New) The method of claim 30, wherein said hormone is follicle stimulating hormone.

33. (New) The method of claim 6, wherein said fertilized avian egg is a chicken egg.

34. (New) The method of claim 6, wherein said primordial germ cells are isolated from said gonads at stage 31-36.

35. (New) The method of claim 6, wherein sex-determined gonads are selected according to morphology.

36. (New) A method of isolating sex-determined PGCs comprising the steps of:
(a) incubating an egg until the gonads sexually differentiate;
(b) harvesting the gonads from the developing embryo;
(c) separating the female gonads from the male gonads based on morphological differences; and

(d) isolating PGCs from female gonads and from male gonads, wherein PGCs isolated from female gonads are segregated from PGCs isolated from male gonads.

37. (New) A method of producing a sex-determined recipient egg comprising contacting a developing egg with a hormone before the embryo sexually differentiates.

38. (New) The method of claim 37, wherein said hormone is testosterone.

39. (New) The method of claim 37, wherein said hormone is estrogen.

40. (New) The method of claim 37, wherein said hormone is follicle stimulating hormone.